

DESCRIPTION	
<b>Species Reactivity</b>	Human/Mouse/Rat
<b>Specificity</b>	Detects human NF-L in direct ELISAs. Detects human, mouse, and rat NF-L in Western blots.
<b>Source</b>	Monoclonal Mouse IgG <sub>1</sub> Clone # 985912
<b>Purification</b>	Protein A or G purified from hybridoma culture supernatant
<b>Immunogen</b>	<i>E. coli</i> -derived recombinant human NF-L Met1-Asp338 Accession # P07196
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

**APPLICATIONS**

Please Note: Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.

	Recommended Concentration	Sample
Dual RNAscope ISH-IHC Compatible	5-25 µg/mL	Formalin-fixed paraffin-embedded tissue sections of human cerebellum.
Western Blot	2 µg/mL	See Below
Immunohistochemistry	5-25 µg/mL	See Below

**DATA**

**Western Blot**

**Detection of Human, Mouse, and Rat NF-L by Western Blot.** Western blot shows lysates of SH-SY5Y human neuroblastoma cell line, mouse brain tissue, and rat brain tissue. PVDF membrane was probed with 2 µg/mL of Mouse Anti-Human NF-L Monoclonal Antibody (Catalog # MAB22161) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for NF-L at approximately 70 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 3.

**Immunohistochemistry**

**NF-L in Human Brain.** NF-L was detected in immersion fixed paraffin-embedded sections of human brain (substantia nigra) using Mouse Anti-Human NF-L Monoclonal Antibody (Catalog # MAB22161) at 5 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Mouse IgG VisUCyte™ HRP Polymer Antibody (Catalog # Catalog # VC001). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to neurons. View our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

**Dual RNAscope ISH-IHC Compatible**

**Detection of NEFL mRNA (red) and protein (green/blue) in Human Cerebellum.** NEFL mRNA (red) and protein (green/blue) was detected in formalin-fixed paraffin-embedded tissue sections of human cerebellum. ACD's Integrated Co-Detection Workflow was performed using ACD RNAscope Probe Hs-NEFL (Catalog # 468671) and mouse anti-human NEFL monoclonal antibody (R&D Systems Catalog #MAB22161) at 5 µg/mL. Tissue was stained using RNAscope® 2.5 HD Detection Reagents-RED (Catalog # 322360), Mouse IgG VisUCyte HRP Polymer Antibody (Catalog # VC001) and RNAscope® 2.5 LS Green Accessory Pack (Catalog #322550). Tissue was counterstained with 50% hematoxylin (purple).

## PREPARATION AND STORAGE

<b>Reconstitution</b>	Reconstitute at 0.5 mg/mL in sterile PBS.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
<b>Stability &amp; Storage</b>	<b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b> <ul style="list-style-type: none"><li>• 12 months from date of receipt, -20 to -70 °C as supplied.</li><li>• 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li><li>• 6 months, -20 to -70 °C under sterile conditions after reconstitution.</li></ul>

## BACKGROUND

NF-L is a 68 kDa light chain cytoskeletal intermediate filament protein that is expressed in neurons. It associates with the 125 kDa NF-M and the 200 kDa NF-H to form neurofilaments.